Chinese Journal of Physiology 61(1): 42-49, 2018
DOI: 10.4077/CJP.2018.BAG523

Modulatory Effect of Concomitant Administration of Insulin and Vanadium on Inflammatory Biomarkers in Type 2 Diabetic Rats: Role of Adiponectin

Ismaeel Bin-Jaliah[1], Hussein F. Sakr[2, 3], Mohamed D. Morsy[1, 4], Mohamed Dallak[1], and Mohamed A. Haidara[1, 5]

[1]Department of Physiology, College of Medicine, King Khalid University, Abha, Aseer, Saudi Arabia
[2]Department of Medical Physiology, Faculty of Medicine and health sciences, Sultan Qaboos University, Muscat, Oman
[3]Department of Medical Physiology, Faculty of Medicine, Mansoura University, Mansoura, Egypt
[4]Department of Clinical Physiology, Faculty of Medicine, Menoufia University, Shebeen Alkoom, Egypt
and
[5]Department of Physiology, Kasr Al-Aini Faculty of Medicine, Cairo University, Cairo, Egypt

Abstract

The aim of this study is to investigate the effect of vanadium and/or insulin on the pro-inflammatory biomarkers in type 2 diabetes mellitus (T2DM) rat model. Sixty male Sprague Dawley rats were divided into six groups (n = 10). Control group, control vanadium group, T2DM group, insulin-treated diabetic group, vanadium-treated diabetic group, and concomitant insulin and vanadium-treated diabetic group. At the end of the experiment, serum glucose, insulin, lipid profile, tumor necrosis factor alpha (TNF-α), interleukin-6 (IL-6), high sensitivity C reactive protein (hs-CRP), intercellular adhesion molecule-1 (ICAM-1), vascular cell adhesion molecule-1 (VCAM-1) and adiponectin were measured. Administration of insulin and/or vanadium significantly decreased in the plasma levels of glucose, lipid profile, TNF-α, IL-6, hs-CRP, ICAM-1, and VCAM-1 with significant increase in adiponectin in comparison to the diabetic group. Concomitant administration of insulin and vanadium significantly improved the above measured parameters compared to either insulin or vanadium treatment. Based on our results we can conclude that administration of both vanadium and insulin reduced the low-grade systemic inflammation in T2DM, through reduction of both pro-inflammatory cytokines and adhesion molecules and increase adiponectin.

Key Words: adiponectin, cytokines, diabetes, insulin, vanadium

Introduction

Type 2 diabetes mellitus (T2DM) is basically characterized by the presence of insulin resistance (IR) due to receptor or post receptor defect (13). IR and hyperglycemia have been accused in the low grade inflammatory status accompanying T2DM (8, 15, 24) through overproduction of advanced glycation end products (12). Thus, T2DM has been categorized as an inflammatory disease (7, 30) associ-

Corresponding author: Prof. Mohamed A. Haidara, Department of Physiology, College of Medicine, King Khalid University, Abha, SA, PO Box (641), 61421, Saudi Arabia. E-mail: haidaram@hotmail.com, Mobile: 00966540733723
Received: March 25, 2017; Revised: June 5, 2017; Accepted: September 19, 2017.
 ISSN : 0304-4920. http://www.cps.org.tw

ated with disturbed plasma levels of several pro-inflammatory cytokines, and adipocytokines including leptin, adiponectin, tumor necrosis factor alpha (TNF-α) and interleukin-6 (IL-6). Disturbances of these inflammatory mediators have been linked to the onset of IR (45) in the peripheral tissues, particularly the skeletal muscle and adipose tissue (5). IR in T2DM enhances adipocytokines upset as a consequence of chronic positive energy balance in adipose tissue and leads to accumulation of excess triglycerides in adipocytes with subsequent macrophage infiltration and inflammatory reaction (2). Several studies reported that interruption of cytokines in T2DM correlates remarkably with increased IR (33, 39) and regulation of the hepatic acute phase reactant C-reactive protein (CRP) (44, 47). IR and hyperglycemia up-regulate the intercellular adhesion molecule-1 (ICAM-1) and vascular cell adhesion molecule-1 (VCAM-1) in the vascular endothelium in T2DM (11). Up-regulation of ICAM and VCAM promote adhesion of leukocytes to the endothelium with subsequent exaggeration of the already established inflammatory processes (1). ICAM-1 not only promotes atherogenesis (4) but also aggravates organ damage (9) through exacerbation of the inflammatory sequels.

IR and chronic hyperglycemia increase the pro-inflammatory and inflammatory mediators, inducing the reduction in insulin sensitivity and tissue glucose uptake, with subsequent exaggerated inflammatory condition and a vicious circle developed. Breaking this circle may block the inflammatory cascade and prevent development of the low-grade inflammatory reaction associated with T2DM.

Vanadyl sulfate (VOSO) (4) and its complexes with several types of ligands have been proposed as useful for treating DM in experimental diabetic animals. On the basis of a mechanistic study, Vanadyl ion and its complexes have been shown to be effective, not only in treating or relieving both types of DM, but also in preventing the onset of DM (34).

The insulin-mimetic potential and anti-diabetic effects of vanadium compounds in rodents and human have been clearly validated. Vanadium is a transitional trace element that is widely dispersed in nature and its administration has been reported to improve diabetes mellitus in humans (29). Vanadium compounds have been shown to mimic insulin in *in vitro* and *in vivo* systems. They exert anti-diabetic effects in rodent models of type 1 and T2DM, as well as in a restricted number of researches in human diabetic subjects. Thus, vanadium compound displays potential use in diabetes therapy. However, treatment of diabetic animals with inorganic vanadium salts has also been associated with some toxic side-effects on liver, kidneys in addition to gastrointestinal discomfort and decreased body weight (BW) gain (36).

The aim of the present study is to investigate the effects of vanadium and or insulin on the pro-inflammatory and adhesion molecules biomarkers in systemic low – grade inflammation in T2DM rat model.

Preliminary results of this study were presented previously in an abstract form at the International Conference of Physiological Sciences that was hosted by the Chinese Association for Physiological Sciences in Beijing, China (18).

Materials and Methods

Chemicals

Streptozotocin (STZ) was purchased from (Sigma Chemical Company, USA), vanadium and sodium thiopental were purchased from (Bio-Chem, Austria), mixtard insulin was ordered from (Novo Nordisk, Denmark).

Experimental Animals

This study was carried out on 60 male Sprague Dawley rats, (150-200 g BW). Animals were fed with standard laboratory chow and water *ad libitum* and housed in the animal house of King Khalid University, College of Medicine under artificial light/dark cycle of 12 h. The animals were divided into six groups (n = 10): control group (C): rats were injected intraperitoneally (i.p.) once with citrate buffer only (0.1 M, pH 4.5), vanadium control group (CV): rats were injected i.p. with buffer as C group and received vanadyl sulfate of 0.64 mmol/kg weight freshly dissolved in 1 ml of distilled water daily through an esophageal tube (46), diabetic group (D): rats received high fat diet for 2 weeks followed by a single i.p. injection of STZ, 50 mg/kg BW (10), insulin-treated diabetic group (DI): rats were made diabetic as in D group and received mixtard insulin subcutaneously in a dose of 0.75 IU/100 gm weight in 0.75 ml volume once daily (41) after 48 h of induction of diabetes; vanadium-treated diabetic group (DV): rats received the same dose of vanadium as in CV group after 48 h of induction of diabetes, combined insulin and vanadium-treated diabetic group (DIV): rats received both vanadium and insulin with the same doses as in groups DV and DI respectively. The daily treatments for the animals were continued for 6 weeks.

Induction of T2DM

DM was induced by a single i.p. injection of

STZ (50 mg/kg BW) in freshly prepared citrate buffer (0.1 M, pH 4.5) (Sigma Chemical Company, USA), while control rats were injected with vehicle buffer alone. DM was verified by measuring blood glucose through tail-neck blood sampling. Rats with non-fasting blood glucose level of ≥20 mM after 48 h of STZ injection were considered to be diabetic (20).

The study period of the experiment was 8 weeks. Although the failure rate for development of diabetes was 15% and the death rate was 5%, we only used diabetic rats in the experiment whose blood glucose level became above 20 mM. The failed cases of rats that died were excluded from the experimental study group from the start. The experiments were conducted in accordance with the ethical guidelines for investigations in laboratory animals and were approved by the ethical committee of the College of Medicine, King Khalid University (REC-2013-02-07).

Biochemical Measurements

Blood Samples

At the end of experimental period, blood samples were collected by cardiac puncture under anesthesia (sodium thiopentone at 40 mg/kg BW) after an overnight fast for 12 h. These blood samples were collected without anticoagulant, left for 10 min, then centrifuged for 10 min at 4000 r/min to obtain serum, which was stored at –20°C until further biochemical analysis for determination of serum glucose, insulin, adiponectin, IL-6, TNF-α, high sensitivity-CRP (hs-CRP), ICAM-1, VCAM-1, triglyceride (TG), total cholesterol (TC), high density lipoprotein cholesterol (HDL-C), low density lipoprotein cholesterol (LDL-C).

Determination of the Serum Adiponectin, TNF-α, IL-6, hs-CRP, ICAM-1 and VCAM-1

Quantitative determination of serum adiponectin was performed using the mouse/rat adiponectin enzyme-linked immunosorbent assay (ELISA) kit (B-Bridge international, Inc.), according to the manufacturer's instructions. ELISA kits for determination of serum levels of hs-CRP, Cat. No. ERC1021-1), serum IL-6 (Cat No.ELR0IL6-001) and serum TNF-α, Cat No. R63635) were purchased from ASSAYPRO, USA. ICAM-1 (Cat. No. SEA548Ra, USCN Life Science Inc., USA), and VCAM-1 (Cat. No. SEA547Ra, USCN Life Science Inc., USA) levels were measured according to the manufacturer's instructions.

Statistical Analysis

The data was expressed as mean ± standard deviation (SD). Data was processed and analyzed using the SPSS version 10.0 (SPSS, Inc., Chicago, Ill., USA). One-way analysis of variance (ANOVA) was done followed by Tukey's *post hoc* test. Pearson correlation statistical analysis was done for detection of a probable significance between two different parameters. Results were considered significant if $P \leq 0.05$.

Results

Assessment of Metabolic Parameters

After treatment with STZ, fasting glucose increased from (94.48 ± 8.58 to 199.6 ± 16.22 mg/dl), fasting insulin (from 6.25 ± 0.31 to 7.63 ± 0.21 uU/mL), homeostatic model assessment (HOMA)-IR (from 1.5 ± 0.14 to 3.6 ± 0.41), TG (from 83.23 ± 9.12 to 175.53 ± 15 mg/dl), TC (from 147.39 ± 12 to 204.32 ± 15 mg/dl) and LDL-C (from 95.85 ± 8.36 to 139.72 ± 8.18 mg/dl) with reduction of HDL-C (from 69.42 ± 7.34 to 24.82 ± 2.37 mg/dl), in comparison to control ($P < 0.05$), suggesting that T2DM was induced successfully (Table 1).

As can be seen from Table 1, treatment with insulin alone cause significant decrease glucose level in comparison to diabetic rats (169.6 ± 14 mg/dl) but not in comparison to control associated with increased insulin (7.44 ± 0.14 uU/mL) and HOMA-IR (2.9 ± 0.32) in comparison to control. Treatment of diabetic rats with vanadium alone decreases glucose level significantly (121.8 ± 12 mg/dl) in comparison to diabetic rats but not to control level. However it associated with significant decrease in insulin (4.49 ± 0.35 uU/mL) and HOMA-IR (1.9 ± 0.21) in comparison to diabetic and control rats, surprisingly data showed decrease insulin level in comparison to control.

Administration of both vanadium and insulin caused significant decreased glucose level (107.28 ± 9.12 mg/dl), insulin (5.30 ± 0.81 uU/mL) and HOMA-IR (1.4 ± 0.18) in comparison to diabetic but does not return to control level.

Table 1 also showed that treatment with insulin or vanadium alone significantly decreases TG, TC and LDL-C associated with increased HDL-C in comparison to diabetic rats but not to control level. Administration of both vanadium and insulin also causes significant decrease of TG, TC and LDL-C associated with increased HDL-C in comparison to diabetic rats but not to control level, though there is decrease in glucose in comparison to diabetic group associated with decrease IR significantly less than diabetic and control group.

Table 1. Effect of insulin and vanadium treatment on fasting glucose, insulin TG, TC, LDL-C and HDL-C in T2DM rats.

	C	CV	D	DI	DV	DIV
Fasting glucose (mg/dl)	94.48 ± 8.58	95.66 ± 5.27	199.60 ± 16.22^{ab}	169.60 ± 13.85^{abc}	121.80 ± 12.76^{abcd}	107.28 ± 9.12^{abcd}
Fasting insulin (uU/mL)	6.25 ± 0.31	6.39 ± 0.15	7.63 ± 0.21^{ab}	7.44 ± 0.14^{ab}	4.49 ± 0.35^{abcd}	5.30 ± 0.81^{abcd}
HOMA-IR	1.50 ± 0.14	1.40 ± 0.15	3.60 ± 0.41^{ab}	2.90 ± 0.32^{ab}	1.90 ± 0.21^{abcd}	1.40 ± 0.18^{cd}
TG (mg/dl)	83.23 ± 9.12	81.38 ± 8.29	175.53 ± 15.31^{ab}	133.67 ± 12.49^{abc}	129.00 ± 11.48^{abc}	104.57 ± 9.87^{cde}
TC (mg/dl)	147.39 ± 12.48	143.47 ± 13.9	204.32 ± 15.8^{ab}	174.30 ± 18.21^{abc}	183.90 ± 17.75^{abc}	153.63 ± 13.4^{cde}
LDL-C (mg/dl)	95.85 ± 8.36	92.41 ± 10.32	139.72 ± 8.18^{ab}	122.34 ± 7.4^{abc}	128.40 ± 9.37^{abc}	107.35 ± 9.39^{cde}
HDL-C (mg/dl)	69.42 ± 7.34	71.94 ± 8.12	24.82 ± 2.37^{ab}	35.31 ± 3.14^{abc}	32.38 ± 3.95^{abc}	55.81 ± 6.72^{cde}

Results were expressed as mean ± SD. C: control, CV: control + vanadium, D: diabetic group, DI: diabetic rats treated with insulin, DV: diabetic rats treated with vanadium, DIV: diabetic rats treated with insulin and vanadium. a, $P < 0.05$ as compared to C; b, $P < 0.05$ as compared to CV; c, $P < 0.05$ as compared to D; d, $P < 0.05$ as compared to DI; e, $P < 0.05$ as compared to DV.

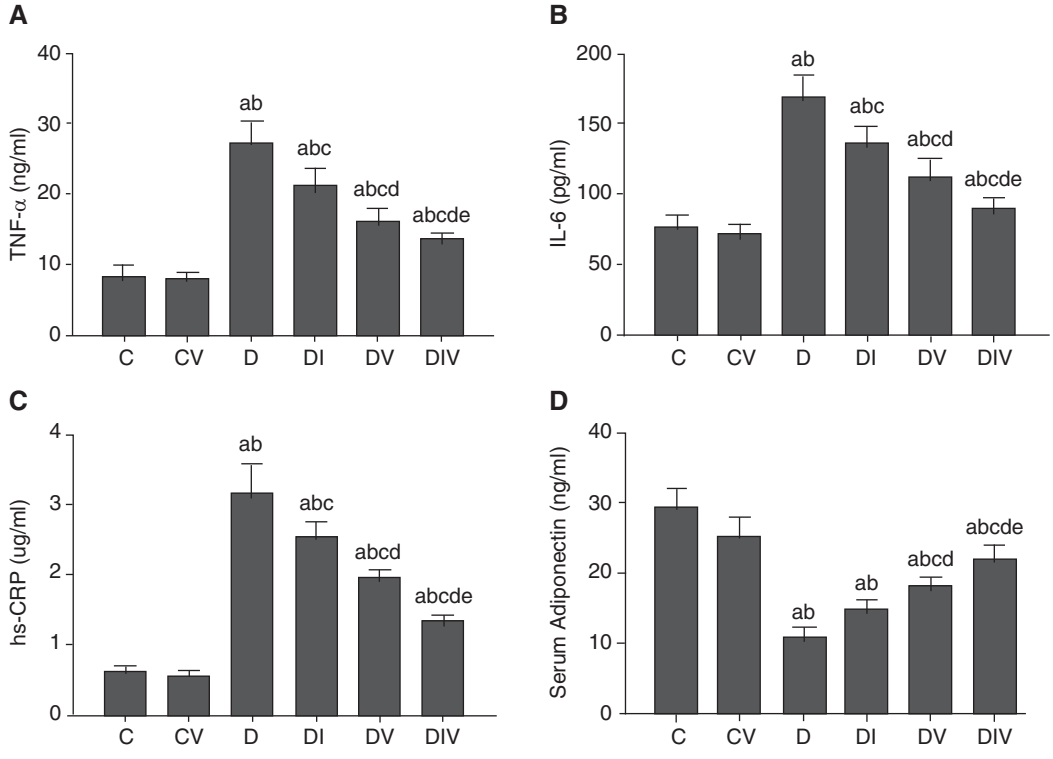

Fig. 1. Serum level of TNF-α (A), IL-6 (B), hs-CRP (C), and adiponectin (D) in the different groups. Results were expressed as mean ± SD of 10 rats. C: control, CV: control + vanadium, D: diabetic group, DI: diabetic rats treated with insulin, DV: diabetic rats treated with vanadium and DIV: diabetic rats treated with insulin and vanadium, a: $P < 0.05$ as compared to C, b: $P < 0.05$ as compared to CV, c: $P < 0.05$ as compared to D; d: $P < 0.05$ as compared to DI; e: $P < 0.05$ as compared to DV.

Effect of Insulin and Vanadium on Pro-Inflammatory Cytokines in T2DM

Figs. 1A, 1B and 1C showed significant increase ($P < 0.05$) of IL-6, TNF-α and hs-CRP in diabetic group as compared to control group. Administration of insulin and or vanadium treatment significantly ($P < 0.05$) reduced the pro-inflamma-

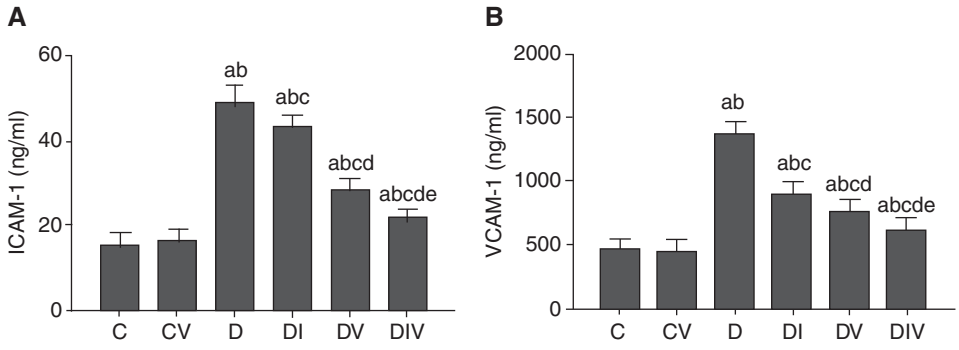

Fig. 2. Serum level of ICAM-1 (A) and VCAM-1 (B) in the different groups. Results were expressed as mean ± SD of 10 rats. C: control, CV: control + vanadium, D: diabetic group, DI: diabetic rats treated with insulin, DV: diabetic rats treated with vanadium and DIV: diabetic rats treated with insulin and vanadium, a: $P < 0.05$ as compared to C; b: $P < 0.05$ as compared to CV; c: $P < 0.05$ as compared to D; d: $P < 0.05$ as compared to DI; e: $P < 0.05$ as compared to DV.

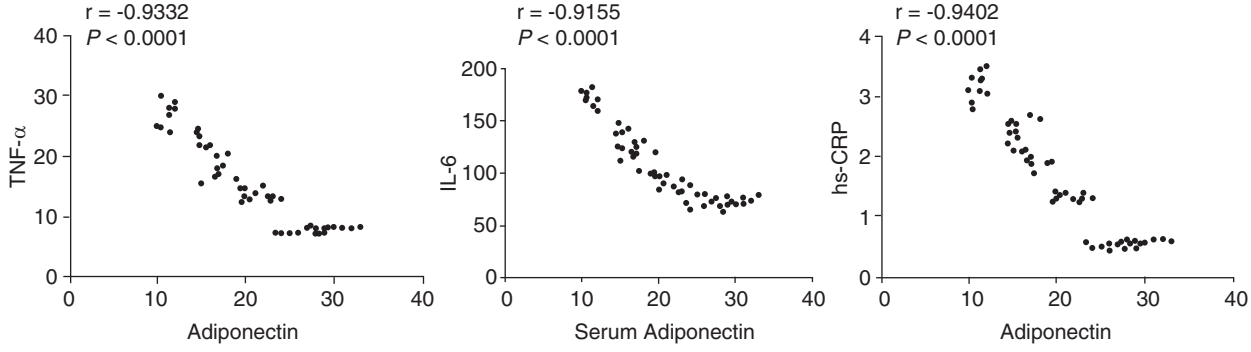

Fig. 3. Negative correlation between adiponectin and TNF-α, for total numbers (n = 60). Negative correlation between adiponectin and IL-6, for total numbers (n = 60). Negative correlation between adiponectin and hs-CRP, for total numbers (n = 60).

tory cytokines in comparison to diabetic group.

Effect of Insulin and Vanadium on Adiponectin Level in T2DM

Adiponectin significantly ($P < 0.05$) decreased in T2DM rats as compared to control. Data obtained from DI, DV and DIV showed that adiponectin increased significantly ($P < 0.05$) as compared to D group. The best ameliorative effect was in DIV group as seen from Fig. 1D.

Effect of Insulin and Vanadium on Serum Adhesion Molecules Levels

Fig. 2A and 2B presents significant increase ($P < 0.05$) of adhesion molecules (ICAM-1 and VCAM-1) as compared to control group. DI, DV and DIV groups showed a significant decrease in ICAM-1 and VCAM-1 as compared to diabetic group.

Correlations

There is significant negative correlation between adiponectin and TNF-α ($P < 0.0001$), (r = -0.9332), significant negative correlation between adiponectin and IL-6 ($P < 0.0001$), (r = -0.9115) and significant negative correlation between adiponectin and hs-CRP ($P < 0.0001$), (r = -0.9402) as shown in Fig. 3.

Discussion

After treatment with STZ, fasting glucose, HOMA-IR, TG, TC and LDL-C associated with reduction of HDL- C, in comparison to control ($P < 0.05$), suggesting that T2DM was induced successfully (Table 1). Administration of insulin and or vanadium modulated the metabolic dysfunction, reduced the pro-inflammatory cytokines and adhesion molecules and increased the circulating adiponectin in T2DM rat model.

Our data showed that administration of vanadium significantly reduced the fasting serum glucose, insulin, TG, TC and LDL-C with significant increase in HDL-C in diabetic rats. These results

are in good agreement with other studies that elaborated the plausible effect of vanadium treatment of T2DM (10, 38). In our results, the levels of fasting glucose decrease in comparison to diabetic rats but did not come back to control level. Insulin level in DV group is significantly lowered than those of C and CV groups and is associated with increased insulin sensitivity. This is in accordance with data that showed that Vanadyl sulfate at maximal tolerated doses for 6 weeks lowers plasma insulin levels and enhances hepatic and muscle insulin sensitivity in T2DM (19). The glucose-lowering effect of vanadium correlated well with the reduction in fasting glucose, but not with insulin-mediated glucose disposal, suggesting that liver, rather than muscle, is the primary target of vanadium action at therapeutic doses in T2DM. Data also showed that administration of vanadium to diabetic rats lowered the required dose of vanadium by 75% (31). Our results also showed that the levels of HOMA-IR and LDL-C of DIV groups were reversed to similar levels of C and CV groups, whereas the levels of fasting glucose, TG and TC of DIV group were still significantly higher than those of C and CV groups which raised an important issue, that further studies are needed to establish the effectiveness of Vanadyl ion in diabetes mellitus. Current research indicated that administration of vanadium to STZ-induced diabetic rats, improved diabetic nephropathy and increased cardiac performance (29).

Previous research has documented that increased pro-inflammatory cytokines levels in diabetics predict the development of atherothrombotic diseases. The inflammatory state is closely related to IR where adipocytes; especially in the obese; secrete a number of pro-inflammatory cytokines (40). Various cytokines directly inhibit insulin signaling (42). Adipocytokines act through key pro-inflammatory regulators such as nuclear factor-κB and the c-Jun NH2-terminal kinase (JNK)/activator protein 1 (AP-1) signaling pathways (14) to modulate the expression of genes coding for many inflammatory proteins and to alter insulin signaling. These actions have two basic consequences: first, to enhance and maintain the pro-inflammatory diathesis, and second, to decrease insulin sensitivity.

Increased levels of circulating inflammatory biomarkers, IL-6, TNF-α and hs-CRP, in T2DM group are broadly consistent with the major trends in previous studies (6, 35) that documented that pro-inflammatory adipokines and cytokines act in an endocrine manner to prompt IR and metabolic derangement (16). Adipokines, including TNF-α, IL-1, resistin and lipocalin-2, possesses pro-inflammatory properties and exerts detrimental effects on cardiac and vascular functions (25).

IR is associated with decreased adiponectin expression (17, 32, 33) through activation of DNA (cytosine-5)-methyltransferase1 (DNMT1). Activated DNMT1 selectively methylates compact chromatin structure in the adiponectin promoter impeding its expression. Suppressing DNMT1 activity with a DNMT inhibitor resulted in the amelioration of IR in an adiponectin-dependent manner (23).

In the present study, administration of insulin and or vanadium to diabetic rats increased serum adiponectin and ameliorated the metabolic dysfunction. In agreement with our data, a recent study showed that vanadium increased adiponectin level in high-fat high-sucrose diet-induced hyperglycemic rats (26). Lu *et al*. concluded that administration of vanadium decrease leptin, resistin and increase adiponectin in induced T2DM rats (27). Additionally, our results showed a significant negative correlation between adiponectin and TNF-α ($r = -0.9332$), IL-6 ($r = -0.9115$) and hs-CRP ($r = -0.9402$).

The results thus obtained are compatible with reports that showed an inverse relationship between TNF-α and adiponectin (21). TNF-α suppresses the expression and secretion of adiponectin from murine and human adipocytes in cell cultures (28). Also increased plasma adiponectin level is associated with decreased IL-6 (3) and hs-CRP (43).

It is well documented that the adhesion molecules are related to inflammatory processes and may be linked to low-grade inflammation in T2DM (22). Our results illustrated that vanadium and or insulin treatment significantly decreased the adhesion molecules (ICAM-1 and VCAM-1) up regulated in the diabetic group. The possible mechanism of the decrease in the expression of adhesion molecules may be attributed to the modulation of the glucose homeostasis and increased adiponectin presented in our study.

Our study presents a novel view of the effect of vanadium on low grade inflammation associated with induced-T2DM rat model. However, a similar role of vanadium in treatment of human diabetes is yet to be established. The slow progress in this direction may be attributed to the gastrointestinal intolerance of the inorganic vanadium salts. However, the availability of organic vanadium compounds with better tolerance may encourage more thorough assessment of these compounds as potential antidiabetic agents for humans (37).

Conclusion

Based on our results it can be concluded that vanadium ameliorates the low grade systemic inflammation in T2DM rat model. This may be attributed to a decrease in pro-inflammatory cytok-

ines and adhesion molecules through increasing adiponectin abrogated in diabetic rats. The findings suggest that this approach could also be used to study the effect of vanadium on other diabetic complications. Therefore, further work will be done on the remaining issues, as part of our project and will be presented in future papers.

Acknowledgments

This work was supported by grants of King Khalid University. KKU-Project No1 (65). We would also like to thank Ms. Yasmeen Haidara, AUC (American University in Cairo) for English editing of our manuscript.

Conflict of Interest

The authors have no conflict of interest to declare.

References

1. Abbas, A.M. and Sakr, H.F. Effect of magnesium sulfate and thyroxine on inflammatory markers in a rat model of hypothyroidism. *Can. J. Physiol. Pharmacol*. 94: 426-432, 2016.
2. Adamczak, M. and Wiecek, A. The adipose tissue as an endocrine organ. *Semin. Nephrol*. 33: 2-13, 2013.
3. Ajuwon, K.M. and Spurlock, M.E. Adiponectin inhibits LPS-induced NF-kappaB activation and IL-6 production and increases PPARgamma2 expression in adipocytes. *Am. J. Physiol. Regul. Integr. Comp. Physiol*. 288: 16-21, 2005.
4. Cybulsky, M.I., Iiyama, K., Li, H., Zhu, S. and Chen, M. A major role for VCAM-1, but not ICAM-1, in early atherosclerosis. *J. Clin. Invest*. 107: 1255-1262, 2001.
5. DeFronzo, R.A. and Tripathy, D. Skeletal muscle insulin resistance is the primary defect in type 2 diabetes. *Diabetes Care* 32: 157-163, 2009.
6. Di Raimondo, D., Tuttolomondo, A., Butta, C., Casuccio, A. and Giarrusso, L. Metabolic and anti-inflammatory effects of a home-based programme of aerobic physical exercise. *Int. J. Clin. Pract.* 67: 1247-1253, 2013.
7. Donath, M.Y. and Shoelson, S.E. Type 2 diabetes as an inflammatory disease. *Nat. Rev. Immunol*. 11: 98-107, 2011.
8. Duncan, B.B., Schmidt, M.I., Pankow, J.S., Ballantyne, C.M. and Couper, D. Low-grade systemic inflammation and the development of type 2 diabetes: the atherosclerosis risk in communities study. *Diabetes* 52: 1799-1805, 2003.
9. Ge, H., Wen, Y., Yang, G. and Betz, A.L. Increased expression of intercellular adhesion molecule-1 in mouse focal cerebral ischemia model. *Chinese Med. J*. 113: 75-79, 2000.
10. Gruzewska, K., Michno, A., Pawelczyk, T. and Bielarczyk, H. Essentiality and toxicity of vanadium supplements in health and pathology. *J. Physiol. Pharmacol*. 65: 603-611, 2014.
11. Gumuslu, E., Cine, N., Gökbayrak, M.E., Mutlu, O., Celikyurt, I.K. and Ulak, G. Exenatide alters gene expression of neural cell adhesion molecule (NCAM), intercellular cell adhesion molecule (ICAM), and vascular cell adhesion molecule (VCAM) in the hippocampus of type 2 diabetic model mice. *Med. Sci. Monit*. 22: 2664-2669, 2016.
12. Haidara, M.A., Khloussy, H., Ammar, H. and Aal Kassem, L.A. Impact of alpha-tocopherol and vitamin C on endothelial markers in rats with streptozotocin-induced diabetes. *Med. Sci. Monit*. 10: BR41-BR46, 2004.
13. Haidara, M.A., Yassin, H.Z., Zakula, Z., Mikhailidis, D.P. and Isenovic, E.R. Diabetes and antioxidants: myth or reality? *Curr. Vasc. Pharmacol*. 8: 661-672, 2010.
14. Hirosumi, J., Tuncman, G., Chang, L., Gorgun, C.Z. and Uysal, K.T. A central role for JNK in obesity and insulin resistance. *Nature* 420: 333-336, 2002.
15. Hotamisligil, G.S. Inflammation and metabolic disorders. *Nature* 444: 860-867, 2006.
16. Hotamisligil, G.S. and Erbay, E. Nutrient sensing and inflammation in metabolic diseases. *Nat. Rev. Immunol*. 8: 923-934, 2008.
17. Hussein, A., Omar, N.M., Sakr, H., Elsamanoudy, A.Z. and Shaheen, D. Modulation of metabolic and cardiac dysfunctions by insulin sensitizers and angiotensin receptor blocker in rat model of type 2 diabetes mellitus. *Can. J. Physiol. Pharmacol*. 89: 216-226, 2011.
18. Bin-Jaliah, I., Haidara, M.A. and Sakr, H.F. Ameliorative effects of concomitant vanadium treatment with insulin on systemic inflammation in a rat model of type 2 diabetes mellitus. *Proc. Int. Conf. Physiol. Sci*. P7-1: 460-461, 2016.
19. Marzban, L. and McNeil, J.H. Insulin-like actions of vanadium: potential as a therapeutic agent. *J. Trace Ele. Exp. Med*. 16: 253-267, 2003.
20. Kedziora-Kornatowska, K.Z., Luciak, M., Blaszczyk, J. and Pawlak, W. Effect of aminoguanidine on erythrocyte lipid peroxidation and activities of antioxidant enzymes in experimental diabetes. *Clin. Chem. Lab. Med*. 36: 771-775, 1998.
21. Kern, P.A., Di Gregorio, G.B., Lu, T., Rassouli, N. and Ranganathan, G. Adiponectin expression from human adipose tissue: relation to obesity, insulin resistance, and tumor necrosis factor-α expression. *Diabetes* 52: 1779-1785, 2003.
22. Khalfaoui, T., Lizard, G. and Ouertani-Meddeb, A. Adhesion molecules (ICAM-1 and VCAM-1) and diabetic retinopathy in type 2 diabetes. *J. Mol. Histol.* 39: 243-149, 2008.
23. Kim, A.Y., Park, Y.J., Pan, X., Shin, K.C. and Kwak, S.H. Obesity-induced DNA hypermethylation of the adiponectin gene mediates insulin resistance. *Nat. Commun*. 6: 7585, 2015.
24. Kolb, H. and Mandrup-Poulsen, T. An immune origin of type 2 diabetes? *Diabetologia* 48: 1038-1050, 2005.
25. Libby, P., Okamoto, Y., Rocha, V.Z. and Folco, E. Inflammation in atherosclerosis: transition from theory to practice. *Circ. J.* 74: 213-220, 2010.
26. Liu, Y., Xu, J., Guo, Y., Xue, Y., Wang, J. and Xue, C. Ameliorative effect of vanadyl(IV)-ascorbate complex on high-fat high-sucrose diet-induced hyperglycemia, insulin resistance, and oxidative stress in mice. *J. Trace Elem. Med. Biol*. 32: 155-161, 2015.
27. Liu, Y., Zhou, Q., Zhao, Y., Wang, Y. and Wang, J. Enrichment, distribution of vanadium-containing protein in vanadium-enriched sea cucumber apostichopus japonicus and the ameliorative effect on insulin resistance. *Biol. Trace Elem. Res*. 171: 167-175, 2016.
28. Maeda, N., Takahashi, M., Funahashi, T., Kihara, S. and Nishizawa, H. PPARgamma ligands increase expression and plasma concentrations of adiponectin, an adipose-derived protein. *Diabetes* 50: 2094-2099, 2001.
29. Morsy, M.D., Abdel-Razek, H.A. and Osman, O.M. Effect of vanadium on renal Na^+, K^+-ATPase activity in diabetic rats: a possible role of leptin. *J. Physiol. Biochem*. 67: 61-69, 2011.
30. Pradhan, A.D., Manson, J.E., Rifai, N., Buring, J.E. and Ridker, P.M. C-reactive protein, interleukin 6, and risk of developing type 2 diabetes mellitus. *JAMA* 286: 327-334, 2001.
31. Ramanadham, S., Cros, G.H., Mongold, J.J., Serrano, J.J. and McNeill, J.H. Enhanced *in vivo* sensitivity of vanadyl-treated diabetic rats to insulin. *Can. J. Physiol. Pharmacol*. 68: 486-491, 1990.

32. Sakr, H.F. Modulation of metabolic and cardiac dysfunctions by swimming in overweight rats on a high cholesterol and fructose diet: possible role of adiponectin. *J. Physiol. Pharmacol*. 64: 231-240, 2013.
33. Sakr, H.F., Al-Hashem, F.H., El-Naby, W.M., Alkhateeb, M.A. and Zaki, M.S. Preventive roles of swimming exercise and pioglitazone treatment on hepatic dysfunction in a rat model of metabolic syndrome. *Can. J. Physiol. Pharmacol*. 92: 162-170, 2014.
34. Sakurai, H. A new concept: the use of vanadium complexes in the treatment of diabetes mellitus. *Chem. Rec*. 2: 237-248, 2002.
35. Smith, D.T., Carr, L.J., Dorozynski, C. and Gomashe, C. Internet-delivered lifestyle physical activity intervention: limited inflammation and antioxidant capacity efficacy in overweight adults. *J. Appl. Physiol*. 106: 49-56, 1985.
36. Srivastava, A.K. Anti-diabetic and toxic effects of vanadium compounds. *Mol. Cell. Biochem*. 206: 177-182, 2000.
37. Srivastava, A.K. and Mehdi, M.Z. Insulino-mimetic and anti-diabetic effects of vanadium compounds. *Diabet. Med*. 22: 2-13, 2005.
38. Thompson, K.H. and Orvig, C. Vanadium in diabetes: 100 years from phase 0 to phase I. *J. Inorg Bioche*m. 100: 1925-1935, 2006.
39. Tilg, H. The role of cytokines in non-alcoholic fatty liver disease. *Dig. Dis*. 28: 179-185, 2010.
40. Trayhurn, P. and Beattie, J.H. Physiological role of adipose tissue: white adipose tissue as an endocrine and secretory organ. *Proc. Nutr. Soc*. 60: 329-339, 2001.
41. Unlucerci, Y., Bekpinar, S., Gurdol, F. and Seferoglu, G. A study on the relationship between homocysteine and diabetic nephropathy in rats. *Pharmacol. Res*. 45: 249-252, 2002.
42. Uysal, K.T., Wiesbrock, S.M., Marino, M.W. and Hotamisligil, G.S. Protection from obesity-induced insulin resistance in mice lacking TNF-α function. *Nature* 389: 610-614, 1997.
43. Venkatesh, B., Hickman, I., Nisbet, J., Cohen, J. and Prins, J. Changes in serum adiponectin concentrations in critical illness: a preliminary investigation. *Crit. Care* 13: 20, 2009.
44. Wellen, K.E. and Hotamisligil, G.S. Inflammation, stress, and diabetes. *J. Clin. Invest*. 115: 1111-1119, 2005.
45. Yudkin, J.S., Stehouwer, C.D., Emeis, J.J. and Coppack, S.W. C-reactive protein in healthy subjects: associations with obesity, insulin resistance, and endothelial dysfunction: a potential role for cytokines originating from adipose tissue? *Arterioscler. Thromb. Vasc. Biol*. 19: 972-978, 1999.
46. Yuen, V.G., Orvig, C. and McNeill, J.H. Comparison of the glucose-lowering properties of vanadyl sulfate and bis(maltolato) oxovanadium(IV) following acute and chronic administration. *Can. J. Physiol. Pharmacol*. 73: 55-64, 1995.
47. Zhang, K., Shen, X., Wu, J., Sakaki, K. and Saunders, T. Endoplasmic reticulum stress activates cleavage of CREBH to induce a systemic inflammatory response. *Cell* 124: 587-599, 2006.